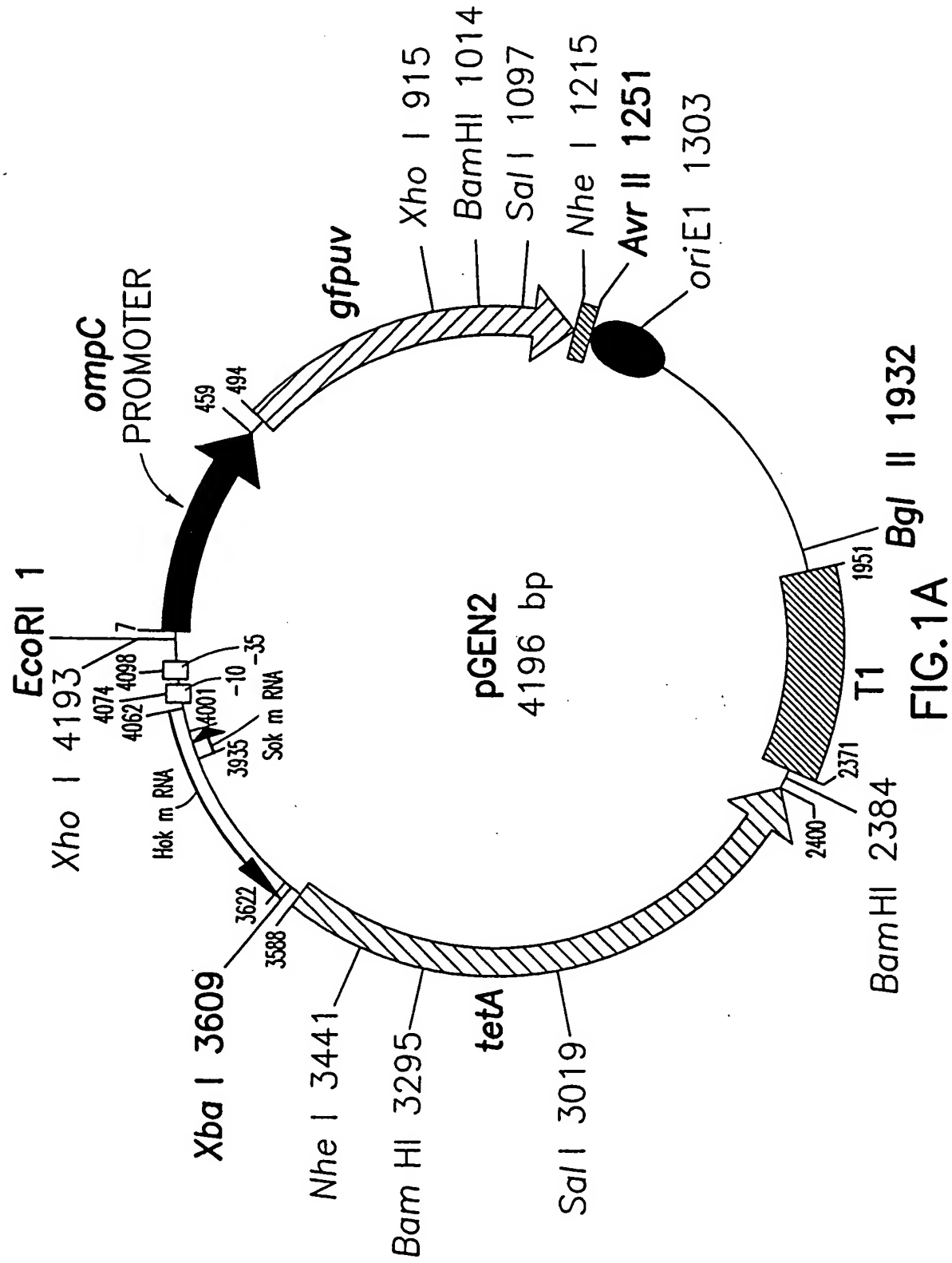

ompC
PROMOTER
gfpuv
Xho I 915
BamHI 1014
Sal I 1097
Nhe I 1215
Avr II 1251
oriE1 1303
EcoRI 1
Xho I 4193
Xba I 3609
Nhe I 3441
Bam HI 3295
tetA
Sal I 3019
BamHI 2384
Bgl II 1932
T1
pGEN2
4196 bp
Hok m RNA
Sok m RNA
-10
-35
7
459
494
4074
4098
4062
4001
3935
3622
3588
2400
2371
1951

FIG.1A

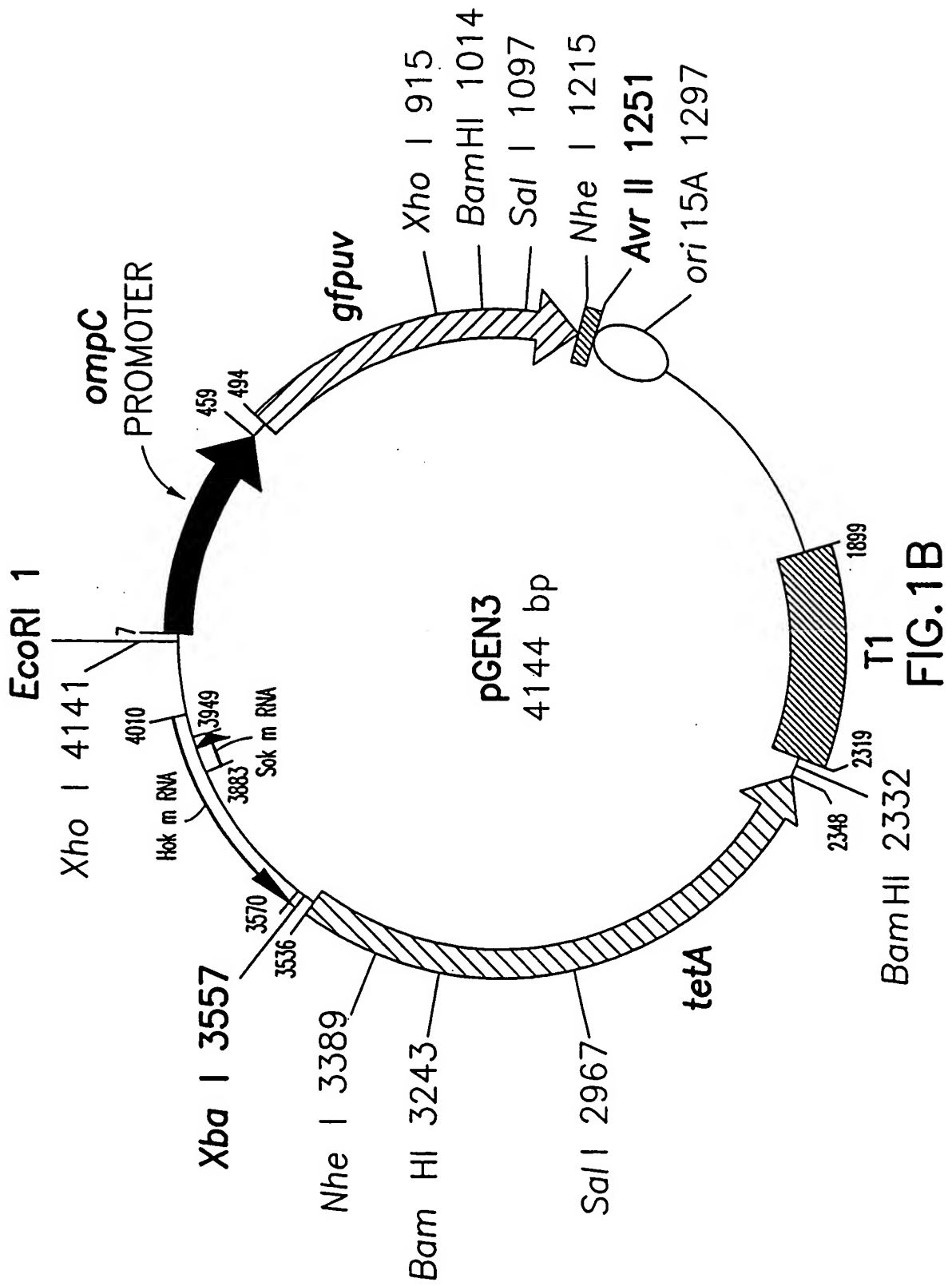

ompC
PROMOTER
gfpuv
Xho I 915
BamHI 1014
Sal I 1097
Nhe I 1215
Avr II 1251
ori 15A 1297
EcoRI 1
Xho I 4141
Xba I 3557
Nhe I 3389
Bam HI 3243
Sal I 2967
tetA
BamHI 2332
T1
pGEN3
4144 bp
7
459
494
1899
2319
2348
3570
3536
4010
3949
3883
Hok m RNA
Sok m RNA

FIG. 1B

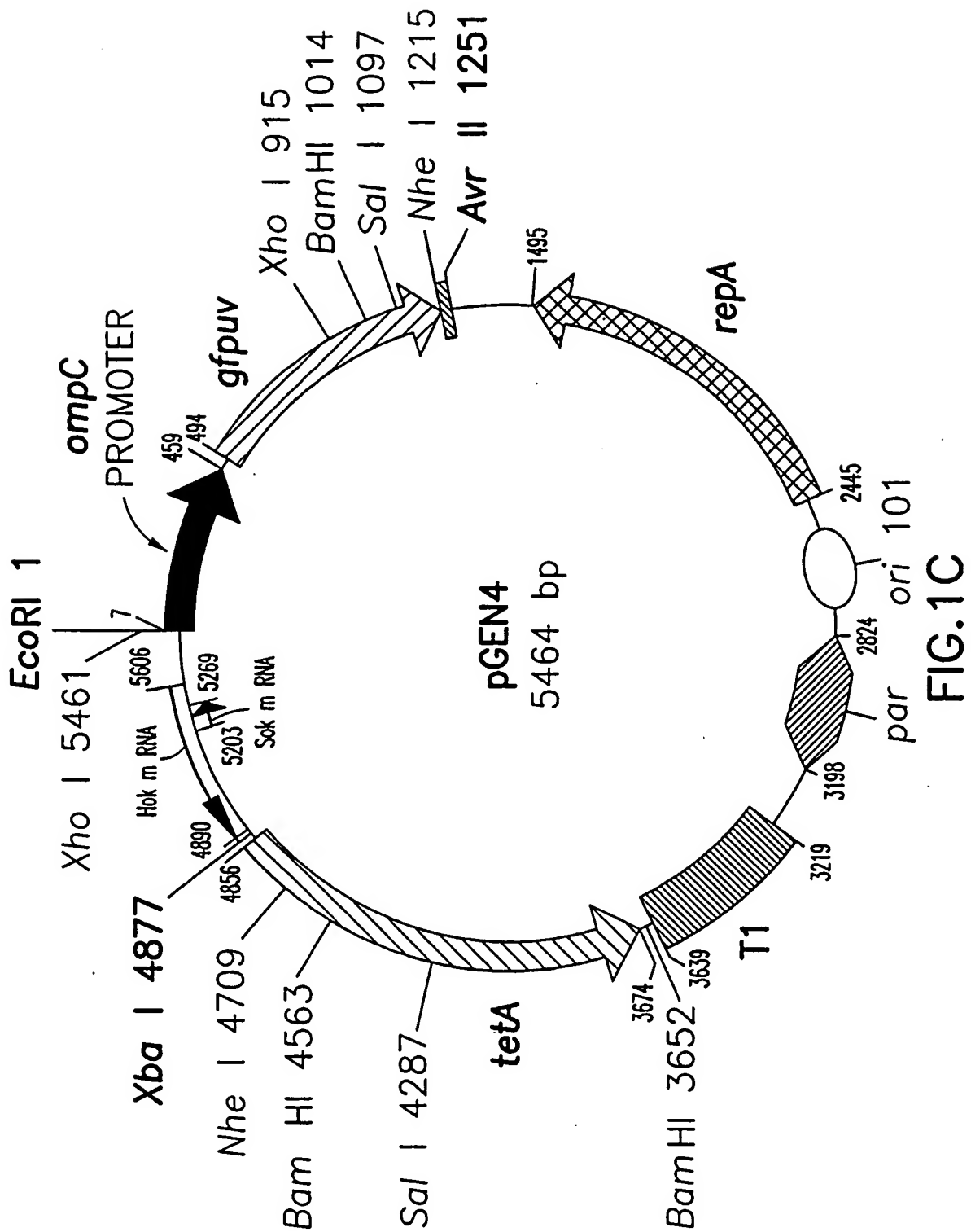

pGEN4
5464 bp
ompC
PROMOTER
gfpuv
repA
par
ori
T1
tetA
EcoRI 1
Xho I 5461
Xba I 4877
Nhe I 4709
Bam HI 4563
Sal I 4287
BamHI 3652
Xho I 915
BamHI 1014
Sal I 1097
Nhe I 1215
Avr II 1251
1495
2445
2824
3198
3219
3639
3674
4856
4890
5203
5269
5606
459
494
7
Hok m RNA
Sok m RNA
101

FIG.1C

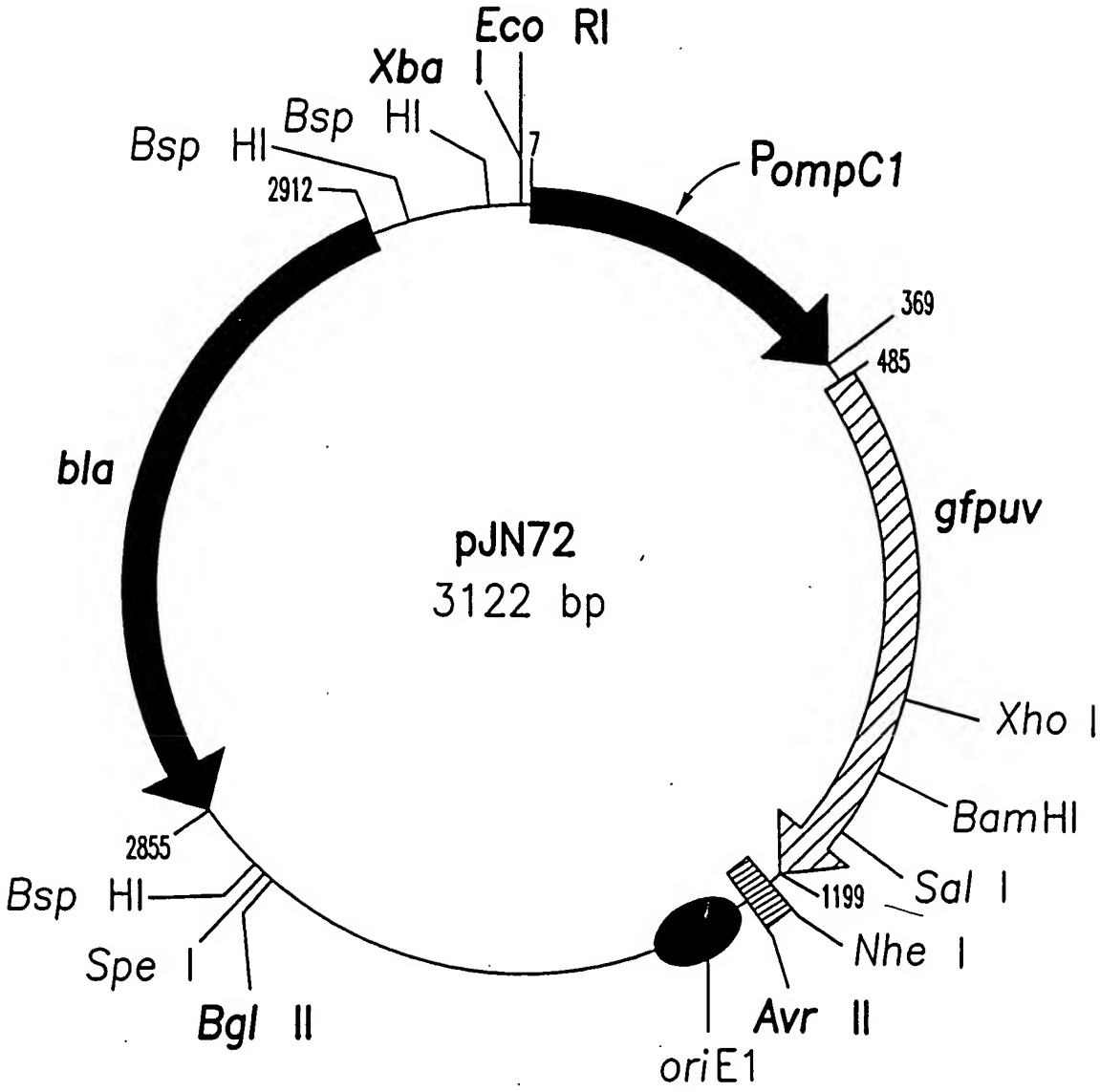

Eco RI
Xba I
Bsp HI
Bsp HI
7
2912
PompC1
369
485
gfpuv
bla
pJN72
3122 bp
Xho I
BamHI
Sal I
1199
Nhe I
Avr II
oriE1
2855
Bsp HI
Spe I
Bgl II

FIG.2A

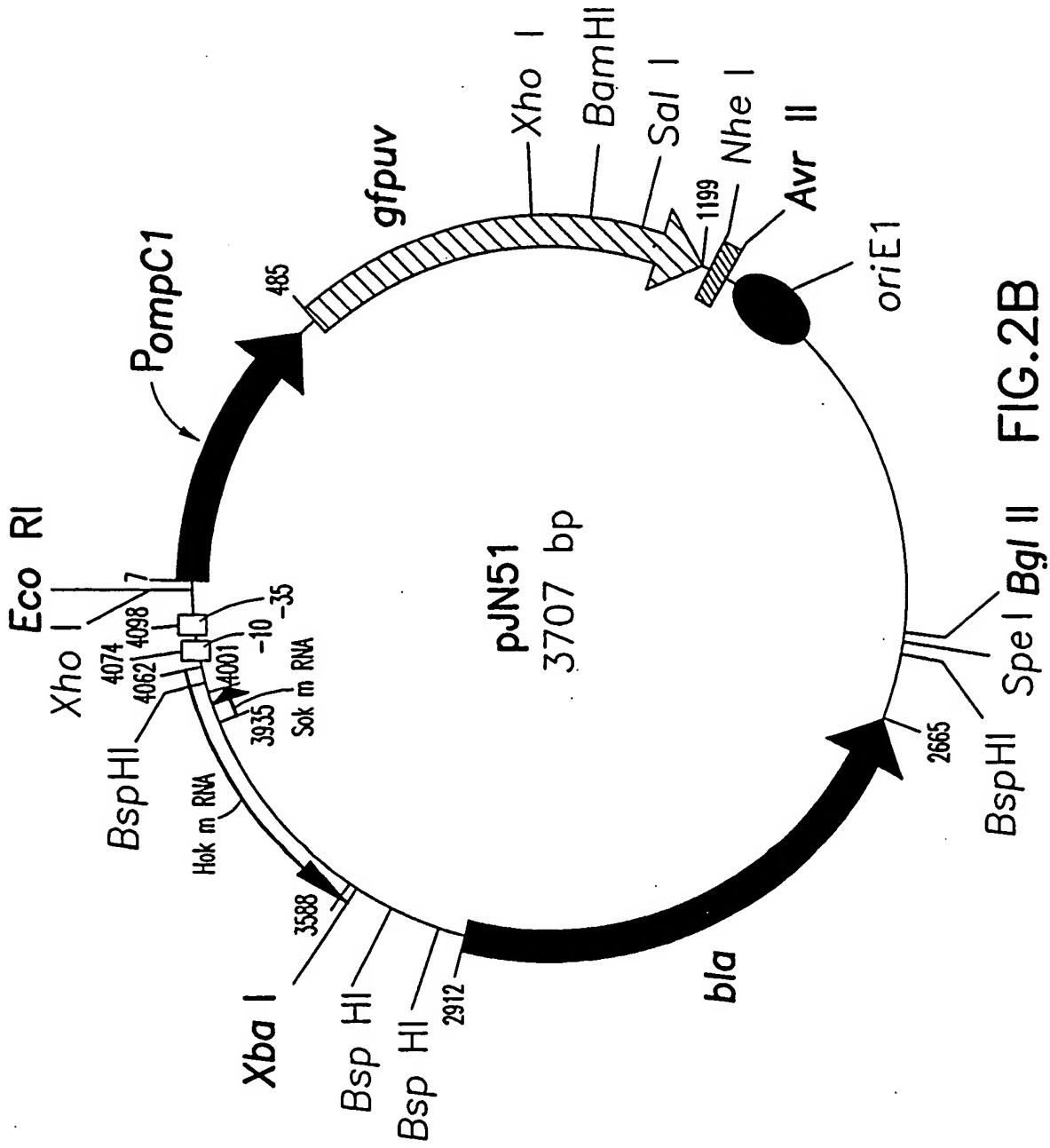

pJN51
3707 bp
PompC1
gfpuv
Xho I
BamHI
Sal I
Nhe I
Avr II
oriE1
Bgl II
Spe I
BspHI
bla
Bsp HI
Bsp HI
Xba I
Hok m RNA
Sok m RNA
-10
-35
Xho I
Eco RI
BspHI
485
1199
2665
2912
3588
3935
4001
4062
4074
4098
7

FIG.2B

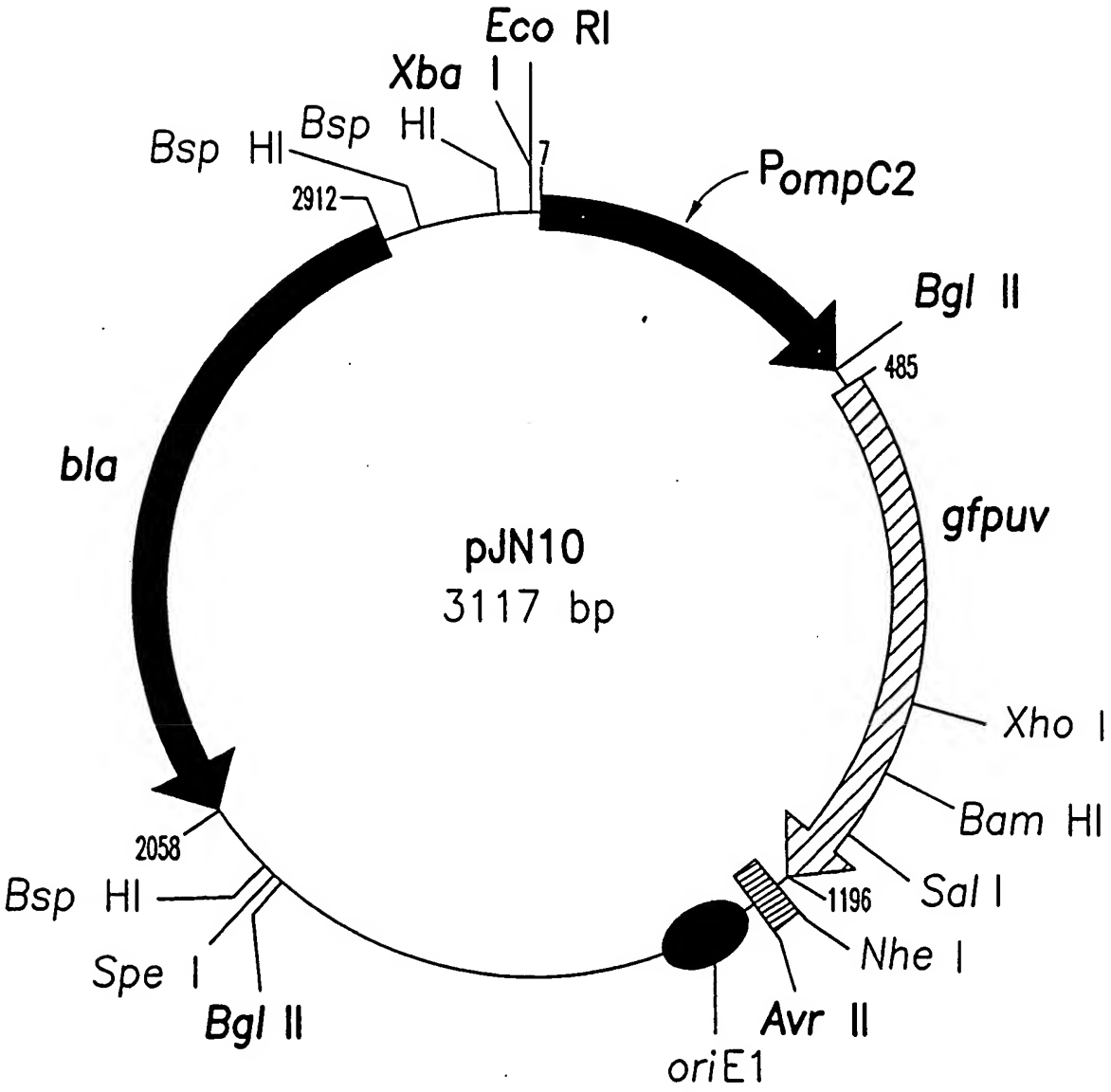

Eco RI
Xba I
Bsp HI
Bsp HI
7
2912
PompC2
Bgl II
485
bla
gfpuv
pJN10
3117 bp
Xho I
Bam HI
Sal I
1196
Nhe I
Avr II
oriE1
2058
Bsp HI
Spe I
Bgl II

FIG.2C

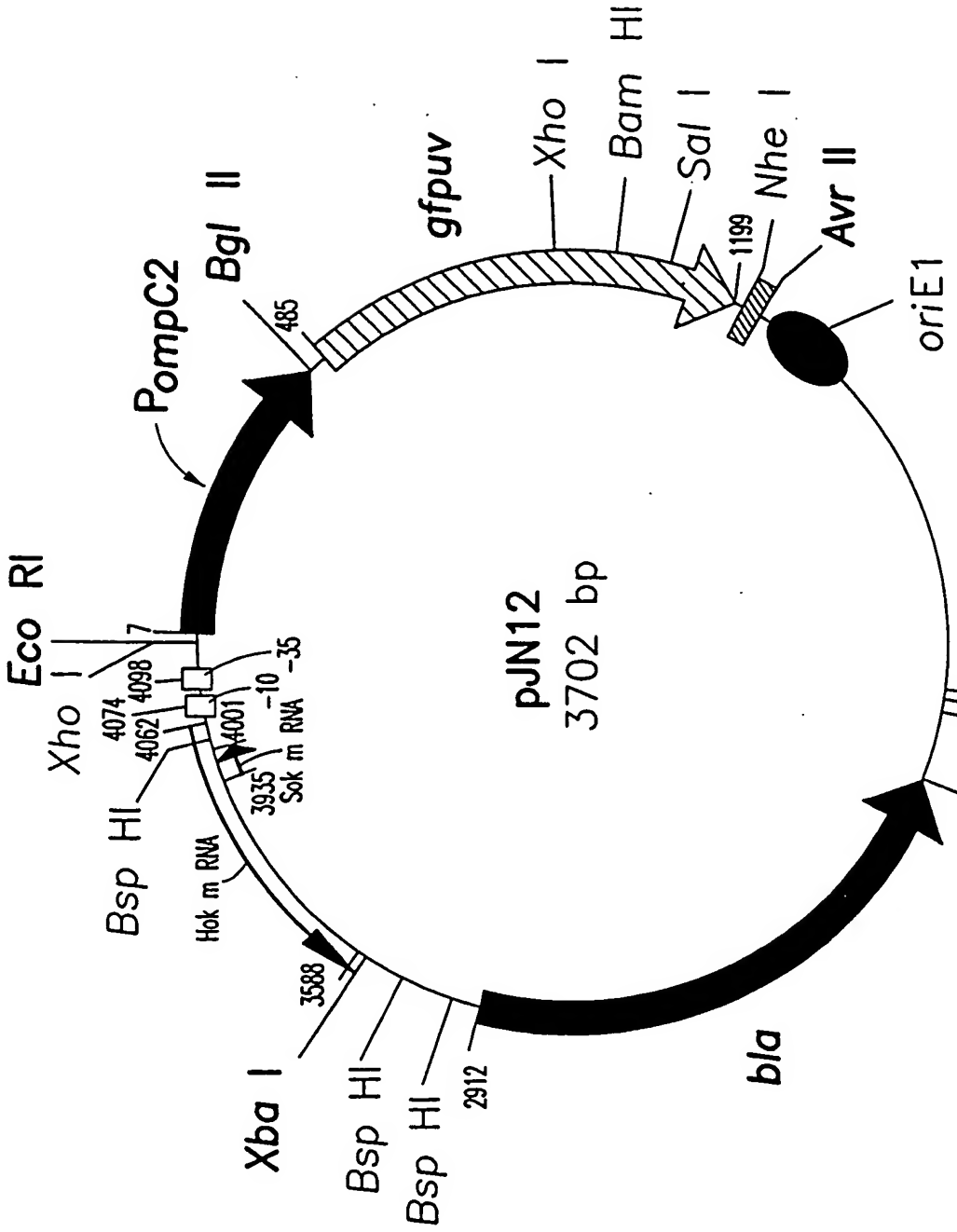

pJN12
3702 bp
PompC2
Bgl II
gfpuv
Xho I
Bam HI
Sal I
Nhe I
Avr II
oriE1
Bgl II
Spe I
Bsp HI
bla
Xba I
Bsp HI
Bsp HI
Hok m RNA
Sok m RNA
Bsp HI
Xho I
Eco RI
485
1199
2055
2912
3588
3935
4001
4062
4074
4098
7
-10
-35

FIG.2D

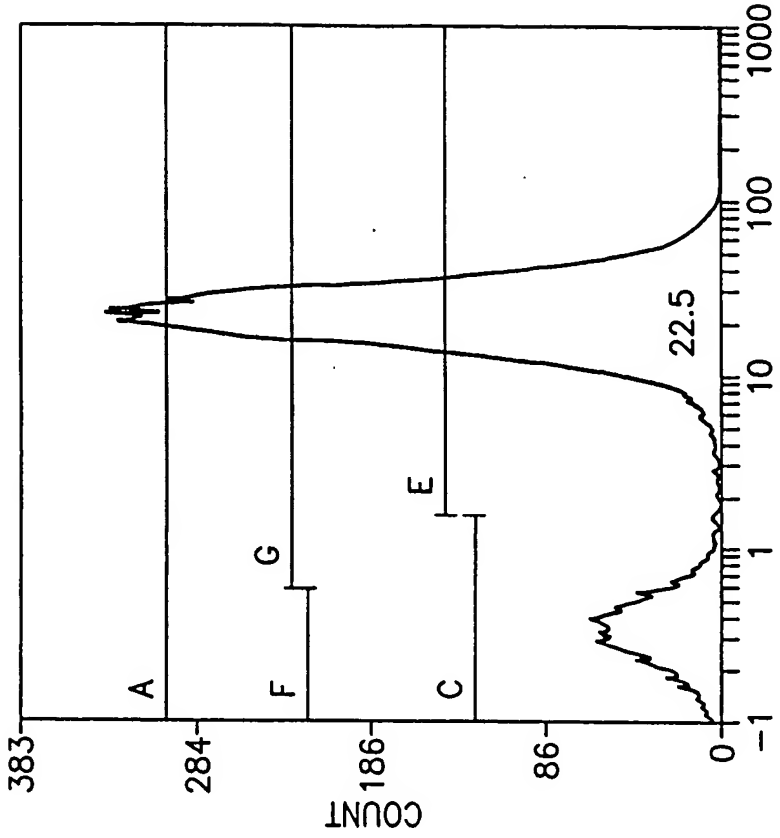

COUNT
383
284
186
86
0
A
F
G
C
E
22.5
−1
1
10
100
1000
GFP

FIG.3B

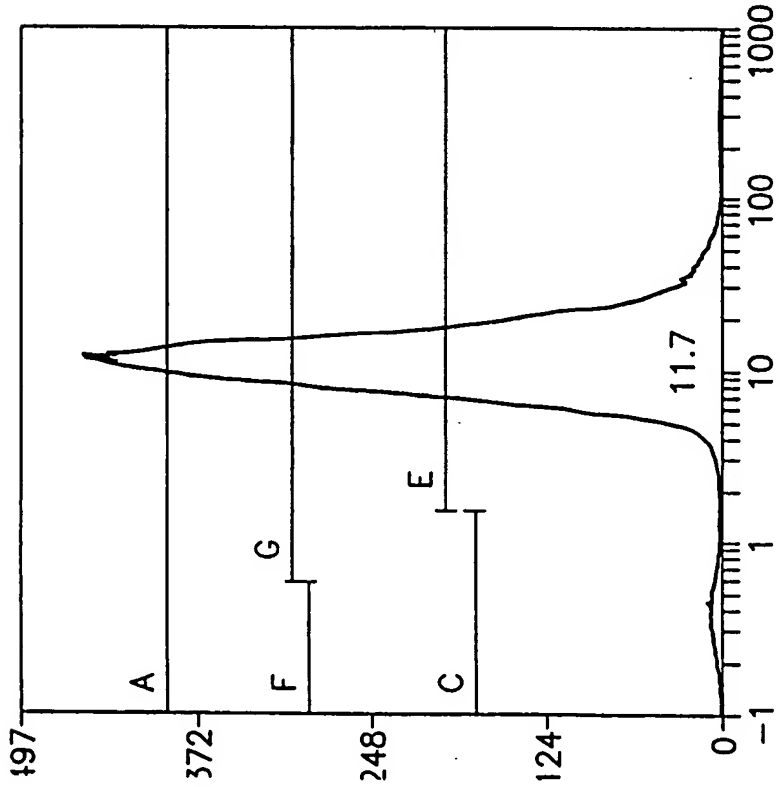

497
372
248
124
0
A
F
G
C
E
11.7
−1
1
10
100
1000
GFP

FIG.3A

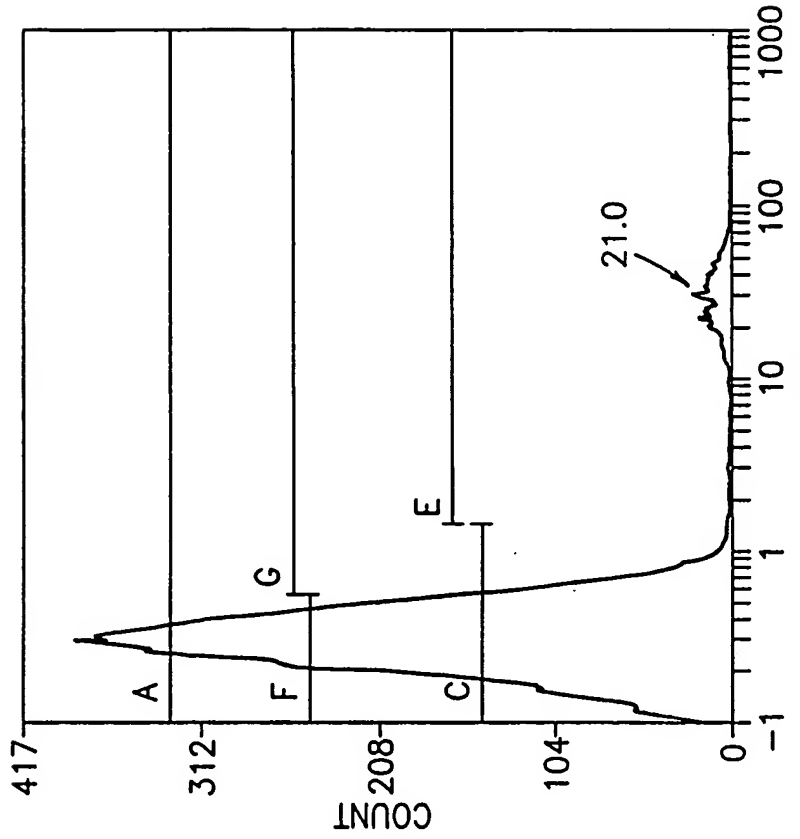

COUNT
417
312
208
104
0
A
F
C
G
E
21.0
−1
1
10
100
1000
GFP

FIG.3D

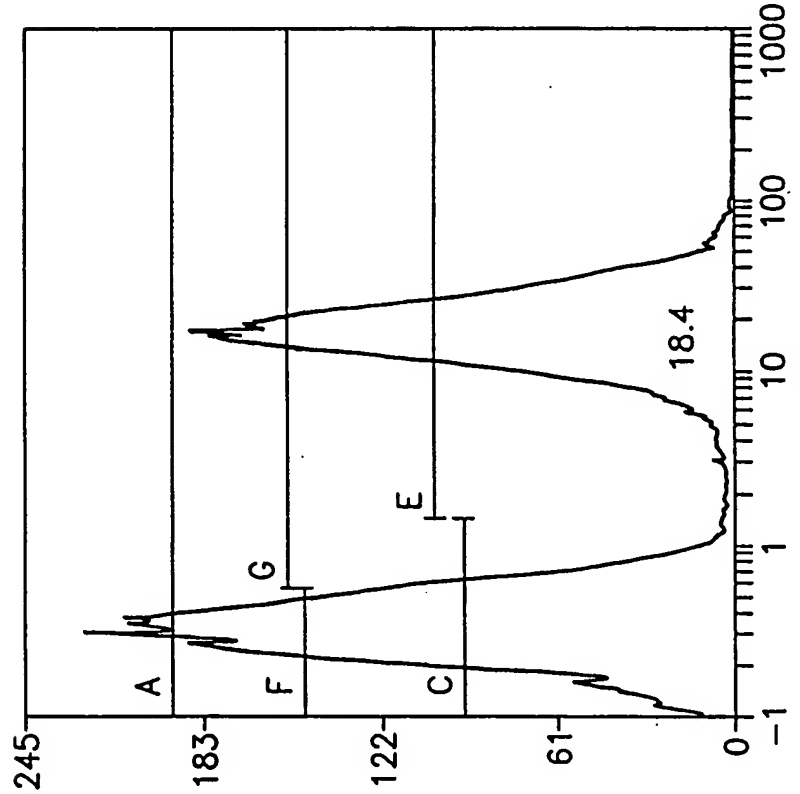

245
183
122
61
0
A
F
C
G
E
18.4
−1
1
10
100
1000
GFP

FIG.3C

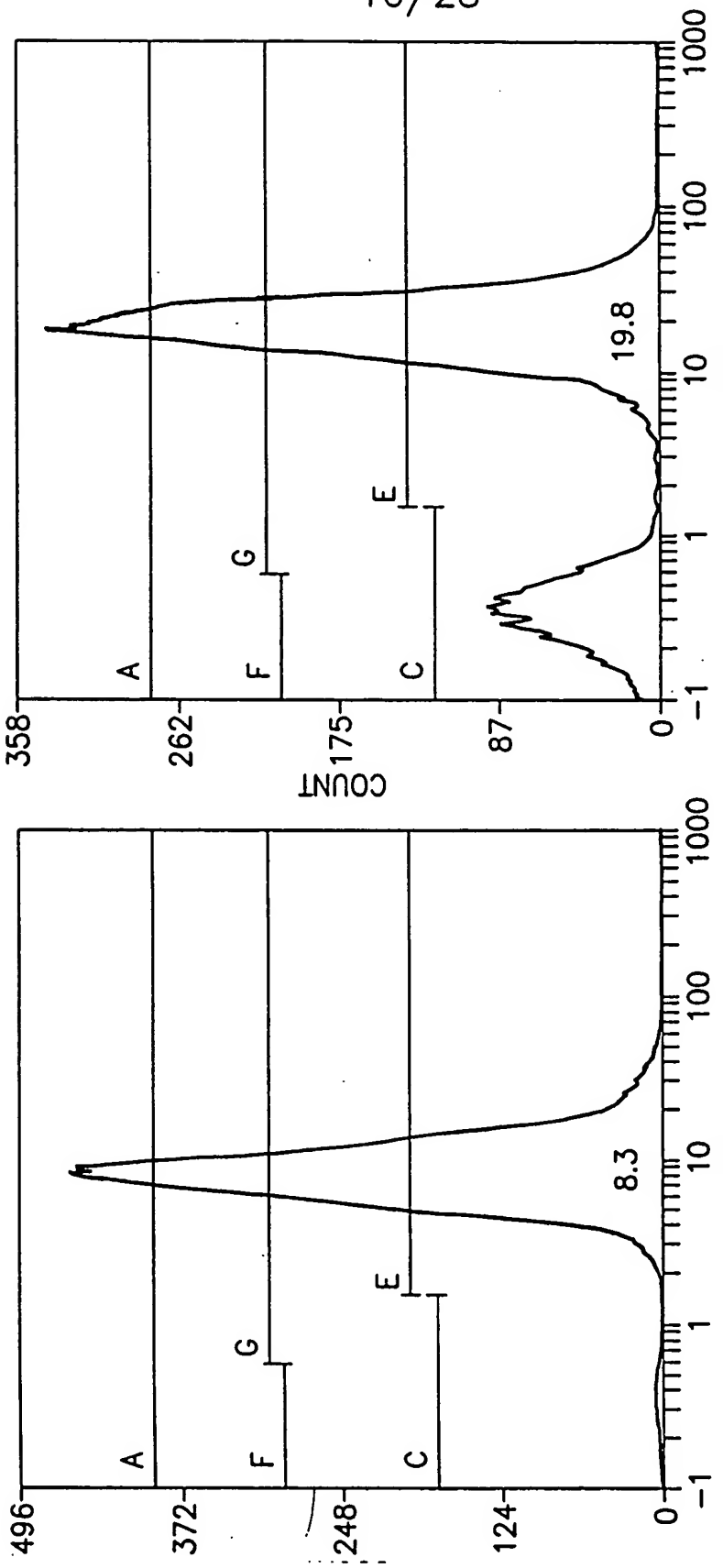


FIG.3F

FIG.3E

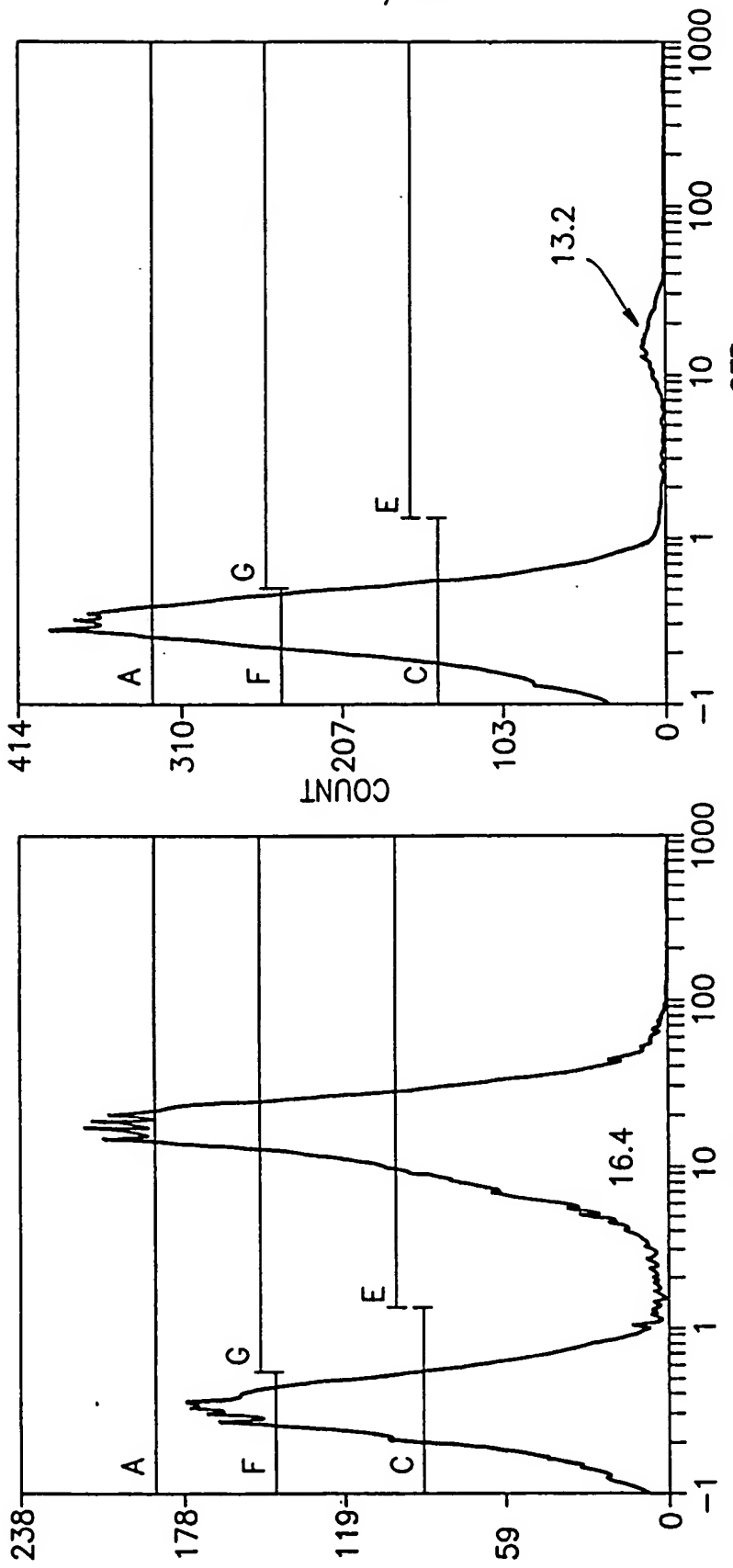

COUNT
414
310
207
103
0
A
F
C
G
E
13.2
1000
100
10
1
−1
GFP
238
178
119
59
0
16.4

FIG.3H

FIG.3G

FIG.4A

```
gaattctgtg gtagcacaga ataatgaaaa gtgtgtaaag aagggtaaaa aaaacogaat 60
gogaggcatc oggttgaaat aggggtaaac agacattcag aaatgaatga oggtaataaa 120
taaagttaat gatgatagog ggagttattc tagttgogag tgaaggtttt gttttgacat 180
tcagtgctgt caaatactta agaataagtt attgatttta accttgaatt attattgctt 240
gatgttaggt gcttatttog ccattcogca ataatcttaa aaagttcoct tgcatttaca 300
ttttgaaaca tctatagoga taaatgaaac atcttaaaag ttttagtatc atattogtgt 360
tggattattc tgcatttttg gggagaatgg acttgoogac tgattaatga gggttaatca 420
gtatgcagtg gcataaaaaa gcaaataaag gcatataaca gatogatctt aaacatccac 480
aggaggatat ctgatgagta aaggagaaga acttttcact ggagttgtcc caattcttgt 540
tgaattagat ggtgatgtta atgggcacaa attttctgtc agtggagagg gtgaaggtga 600
tgcaacatac ggaaaactta occttaaatt tatttgcact actggaaaac tacctgttcc 660
atggccaaca cttgtcacta ctttctctta tggtgttcaa tgcttttccc gttatccgga 720
tcatatgaaa oggcatgact ttttcaagag tgccatgccc gaaggttatg tacaggaacg 780
cactatatct ttcaaagatg acgggaacta caagacgcgt gctgaagtca agtttgaagg 840
tgataccctt gttaatogta togagttaaa aggtattgat tttaaagaag atggaaacat 900
tctoggacac aaactogagt acaactataa ctcacacaat gtatacatca oggcagacaa 960
acaaaagaat ggaatcaaag ctaacttcaa aattogccac aacattgaag atggatcogt 1020
tcaactagca gaccattatc aacaaaatac tccaattggc gatggccctg tccttttacc 1080
```

gacaaccat tacctgtcga cacaatctgc cctttcgaaa gatcccaacg aaaagcgtga 1140
cacatggtc cttcttgagt ttgtaactgc tgctgggatt acacatggca tggatgagct 1200
cacaaataa tgagctagcc cgcctaatga gcgggctttt ttttctcggc ctagggccag 1260
aaaaggcca ggaaccgtaa aaaggccgcg ttgctggcgt ttttccatag gctccgcccc 1320
ctgacgagc atcacaaaaa tcgacgctca agtcagaggt ggcgaaaccc gacaggacta 1380
aaagatacc aggcgtttcc cctggaagc tccctcgtgc gctctcctgt tccgaccctg 1440
cgcttaccg gatacctgtc cgcctttctc ccttcgggaa gcgtggcgct ttctcatagc 1500
cacgctgta ggtatctcag ttcggtgtag gtcgttcgct ccaagctggg ctgtgtgcac 1560
aaccccccg ttcagcccga ccgctgcgcc ttatccggta actatcgtct tgagtccaac 1620
cggtaagac acgacttatc gccactggca gcagccactg gtaacaggat tagcagagcg 1680
ggtatgtag gcggtgctac agagttcttg aagtggtggc ctaactacgg ctacactaga 1740
ggacagtat ttggtatctg cgctctgctg aagccagtta ccttcggaaa aagagttggt 1800
gctcttgat ccggcaaaca aaccaccgct ggtagcggtg gttttttgt ttgcaagcag 1860
agattacgc gcagaaaaaa aggatctcaa gaagatcctt tgatcttttc tacggggtct 1920
acgctcagt agatctaaaa cactaggccc aagagtttgt agaaacgcaa aaaggccatc 1980
gtcaggatg gccttctgct taatttgatg cctggcagtt tatggcgggc gtcctgcccg 2040
caccctccg ggccgttgct tcgcaacgtt caaatccgct cccggcggat ttgtcctact 2100
aggagagcg ttcaccgaca aacaacagat aaaacgaaag gcccagtctt tcgactgagc 2160

FIG.4B

James E. GALEN
PLASMID MAINTENANCE...
A-8703

FIG.4C

```
ctttcgtttt atttgatgcc tggcagttcc ctactctcgc atggggagac cccacactac 2220
catcggcgct acggcgtttc acttctgagt tcggcatggg gtcaggtggg accaccgcgc 2280
tactgccgcc aggcaaattc tgttttatca gaccgcttct gcgttctgat ttaatctgta 2340
tcaggctgaa aatcttctct catccgccaa aacagccaag ctggatcccc gatcttatca 2400
ggtcgaggtg gcccggctcc atgcaccgcg acgcaacgcg gggaggcaga caaggtatag 2460
ggcggcgcct acaatccatg ccaacccgtt ccatgtgctc gccgaggcgg cataaatcgc 2520
cgtgacgatc agcggtccag tgatcgaagt taggctggta agagccgcga gcgatccttg 2580
aagctgtccc tgatggtcgt catctacctg cctggacagc atggcctgca acgcgggcat 2640
cccgatgccg ccggaagcga gaagaatcat aatggggaag gccatccagc ctcgcgtcgc 2700
gaacgccagc aagacgtagc ccagcgcgtc ggccgccatg cccgcgataa tggcctgctt 2760
ctgccgaaa cgtttggtgg cgggaccagt gacgaaggct tgagcgaggg cgtgcaagat 2820
tccgaatacc gcaagcgaca ggccgatcat cgtcgcgctc cagcgaaagc ggtcctgcgc 2880
gaaaatgacc cagagcgctg ccggcacctg tcctacgagt tgcatgataa agaagacagt 2940
cataagtgcg gcgacgatag tcatgccccg cgcccaccgg aaggagctga ctgggttgaa 3000
ggctctcaag ggcatcggtc gacgctctcc cttatgcgac tcctgcatta ggaagcagcc 3060
cagtagtagg ttgaggccgt tgagcaccgc cgccgcaagg aatggtgcat gcaaggagat 3120
ggcgcccaac agtccccccgg ccacggggcc tgccaccata cccacgccga aacaagcgct 3180
catgagcccg aagtggcgag cccgatcttc cccatcggtg atgtcggcga tataggcgcc 3240
```

gcaacogca cctgtggcgc cggtgatgcc ggccacgatg cgtccggcgt agaggatcca 3300
aggacgggt gtggtcgcca tgatcgcgta gtcgatagtg gctccaagta gcgaagcgag 3360
aggactggg cggcggccaa agcggtcgga cagtgctccg agaacgggtg cgcatagaaa 3420
cgcatcaac gcatatagcg ctagcagcac gccatagtga ctggcgatgc tgtcggaatg 3480
acgatatcc cgcaagaggc ccggcagtac cggcataacc aagcctatgc ctacagcatc 3540
agggtgacg gtgccgagga tgacgatgag cgcattgtta gatttcattt ttttttcctc 3600
ctattttct agacaacatc agcaaggaga aagggctac cggcgaacca gcagcccctt 3660
ataaaggcg cttcagtagt cagaccagca tcagtcctga aaaggcgggc ctgcgccgc 3720
ccaggttg ctacttaccg gattcgtaag ccatgaaagc cgccacctcc ctgtgtccgt 3780
cctgtaacg aatctcgcac agcgattttc gtgtcagata agtgaatatc aacagtgtga 3840
acacacgat caacacacac cagacaaggg aacttcgtgg tagtttcatg gccttcttct 3900
cttgcgcaa agcgcggtaa gaggctatcc tgatgtggac tagacatagg gatgcctcgt 3960
gtggttaat gaaaattaac ttactacggg gctatcttct ttctgccaca caacacggca 4020
caaaccacc ttcacgtcat gaggcagaaa gcctcaagcg ccgggcacat catagcccat 4080
tacctgcac gctgaccaca ctcactttcc ctgaaaataa tccgctcatt cagacgttc 4140
cgggaaatc cgtgtgattg ttgccgcatc acgctgcctc ccggagtttg tctcga 4196

FIG.4D

```
ctacaaataa tgagctagcc cgcctaatga gcgggctttt ttttctggc ctaggagata 60
cttaacaggg aagtgagagg gccgcggcaa agccgttttt ccataggctc cgccccctg 120
acaagcatca cgaaatctga cgctcaaatc agtggtggcg aaacccgaca ggactataaa 180
gataccaggc gtttcccct ggcggctccc tcgtgcgctc tcctgttcct gcctttcggt 240
ttacggtgt cattccgctg ttatggccgc gtttgtctca ttccacgcct gacactcagt 300
tccgggtagg cagttcgctc caagctggac tgtatgcacg aaccccccgt tcagtccgac 360
cgctgcgcct tatccggtaa ctatcgtctt gagtccaacc cggaaagaca tgcaaaagca 420
cactggcag cagccactgg taattgattt agaggagtta gtcttgaagt catgcgccgg 480
ttaaggctaa actgaaagga caagttttgg tgactgcgct cctccaagcc agttacctcg 540
gttcaaagag ttggtagctc agagaacctt cgaaaaaccg ccctgcaagg cggtttttc 600
```

FIG.5A

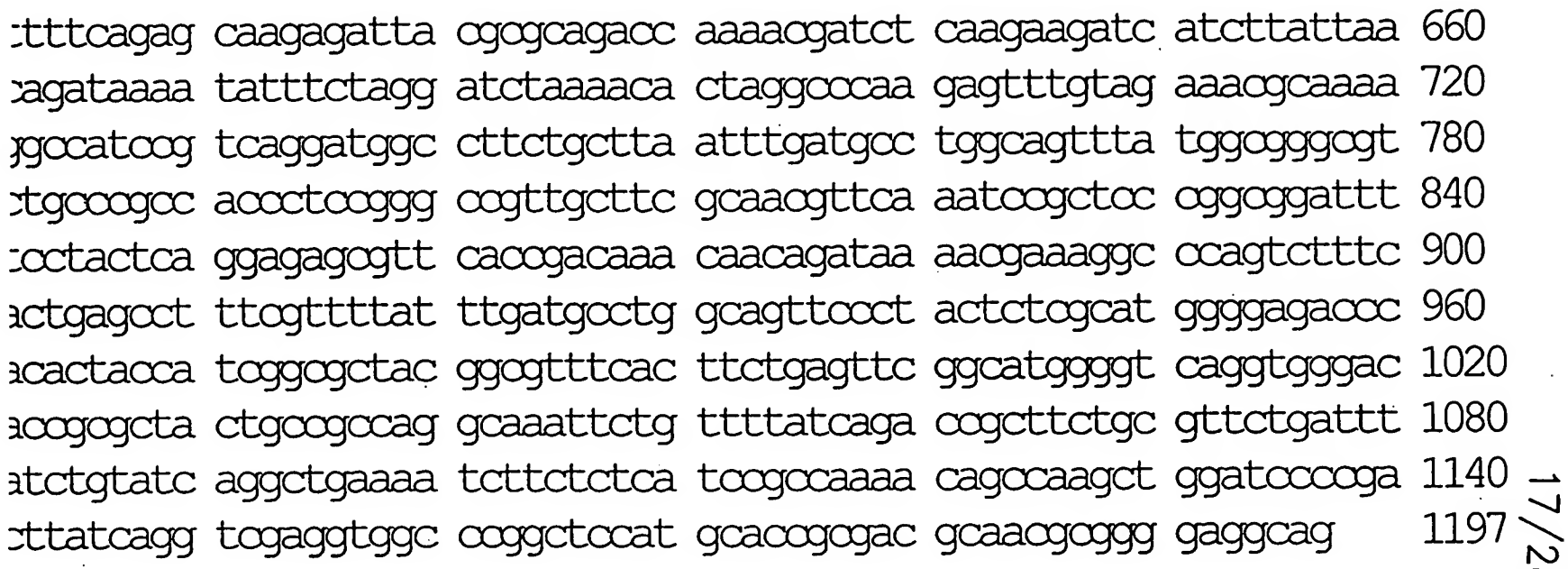

:tttcagag caagagatta cgcgcagacc aaaacgatct caagaagatc atcttattaa 660
:agataaaa tatttctagg atctaaaaca ctaggcccaa gagtttgtag aaacgcaaaa 720
ggccatccg tcaggatggc cttctgctta atttgatgcc tggcagttta tggcgggcgt 780
:tgcccgcc accctccggg ccgttgcttc gcaacgttca aatccgctcc cggcggattt 840
:cctactca ggagagcgtt caccgacaaa caacagataa aacgaaaggc ccagtctttc 900
actgagcct ttcgttttat ttgatgcctg gcagttccct actctcgcat gggggagacc 960
acactacca tcggcgctac ggcgtttcac ttctgagttc ggcatggggt caggtgggac 1020
accgcgcta ctgccgccag gcaaattctg ttttatcaga ccgcttctgc gttctgattt 1080
atctgtatc aggctgaaaa tcttctctca tccgccaaaa cagccaagct ggatcccgga 1140
:ttatcagg tgaggtggc ccggctccat gcaccgcgac gcaacgcggg gaggcag 1197

FIG.5B

ctacaaataa tgagctagcc cgcctaatga gcgggctttt ttttctcggc ctaggtttca 60
cctgttctat taggtgttac atgctgttca tctgttacat tgtcgatctg ttcatggtga 120
acagctttaa atgcaccaaa aactcgtaaa agctctgatg tatctatctt ttttacaccg 180
ttttcatctg tgcatatgga cagttttccc tttgatatct aacggtgaac agttgttcta 240
cttttgtttg ttagtcttga tgcttcactg atagatacaa gagccataag aacctcagat 300
ccttccgtat ttagccagta tgttctctag tgtggttcgt tgtttttgcg tgagccatga 360
gaacgaacca ttgagatcat gcttactttg catgtcactc aaaaattttg cctcaaaact 420
ggtgagctga atttttgcag ttaaagcatc gtgtagtgtt tttcttagtc cgttacgtag 480
gtaggaatct gatgtaatgg ttgttggtat tttgtcacca ttcattttta tctggttgtt 540
ctcaagttcg gttacgagat ccatttgtct atctagttca acttggaaaa tcaacgtatc 600
agtcgggcgg cctcgcttat caaccaccaa tttcatattg ctgtaagtgt ttaaatcttt 660
acttattggt ttcaaaaccc attggttaag ccttttaaac tcatggtagt tattttcaag 720
cattaacatg aacttaaatt catcaaggct aatctctata tttgccttgt gagttttctt 780
ttgtgttagt tcttttaata accactcata aatcctcata gagtatttgt tttcaaaaga 840
cttaacatgt tccagattat attttatgaa ttttttttaac tggaaaagat aaggcaatat 900
ctcttcacta aaaactaatt ctaatttttc gcttgagaac ttggcatagt ttgtccactg 960
gaaaatctca aagcctttaa ccaaaggatt cctgatttcc acagttctcg tcatcagctc 1020
tctggttgct ttagctaata caccataagc attttcccta ctgatgttca tcatctgagc 1080

FIG.6A

James E. GALEN
PLASMID MAINTENANCE...
A-8703

cattggtta taagtgaacg atacogtcog ttctttcctt gtagggtttt caatogtggg 1140
ctgagtagt gccacacagc ataaaattag cttggtttca tgctcogtta agtcatagcg 1200
ctaatogct agttcatttg ctttgaaaac aactaattca gacatacatc tcaattggtc 1260
aggtgattt taatcactat accaattgag atgggctagt caatgataat tactagtcct 1320
ctcctttga gttgtgggta tctgtaaatt ctgctagacc tttgctggaa aacttgtaaa 1380
cctgctaga ccctctgtaa attcogctag acctttgtgt gttttttttg tttatattca 1440
gtggttata atttatagaa taaagaaaga ataaaaaaag ataaaaagaa tagatcccag 1500
cctgtgtat aactcactac tttagtcagt tcogcagtat tacaaaagga tgtogcaaac 1560
ctgtttgct cctctacaaa acagacctta aaacoctaaa ggcttaagta gcaccctogc 1620
agctogggc aaatogctga atattccttt tgtctcogac catcaggcac ctgagtogct 1680
tcttttcog tgacattcag ttcgctgcgc tcacggctct ggcagtgaat gggggtaaat 1740
gcactacag gcgcctttta tggattcatg caaggaaact acccataata caagaaaagc 1800
cgtcacggg cttctcaggg cgttttatgg cgggtctgct atgtggtgct atctgacttt 1860
tgctgttca gcagttcctg ccctctgatt ttccagtctg accacttcgg attatccogt 1920

FIG.6B

gacaggtcat tcagactggc taatgcaccc agtaaggcag cggtatcatc aacaggctta 1980
ccogtcttac tgtcaaccgg atctaaaaca ctaggcccaa gagtttgtag aaacgcaaaa 2040
aggccatccg tcaggatggc cttctgctta atttgatgcc tggcagttta tggcgggcgt 2100
cctgcccgcc accctccggg ccgttgcttc gcaacgttca aatccgctcc cggcggattt 2160
gtcctactca ggagagcgtt caccgacaaa caacagataa aacgaaaggc ccagtctttc 2220
gactgagcct ttcgttttat ttgatgcctg gcagttccct actctgcat gggggagacc 2280
cacactacca tcggcgctac ggcgtttcac ttctgagttc ggcatggggt caggtgggac 2340
cacccgcgcta ctgccgccag gcaaattctg ttttatcaga ccgcttctgc gttctgattt 2400
aatctgtatc aggctgaaaa tcttctctca tccgccaaaa cagccaagct ggatcccccga 2460
tcttatcagg tcgaggtggc ccggctccat gcaccgcgac gcaacgcggg gaggcagaca 2520
aggtataggg cggcgcctac aatccatgcc aacccgttcc atgtgctcgc cgaggcggca 2580
taaatcgccg tgacgatcag cggtccagtg atcgaagtta ggctggtaag agccgcgagc 2640
gatcctt 2647

FIG.6C

James E. GALEN
PLASMID MAINTENANCE...
A-8703

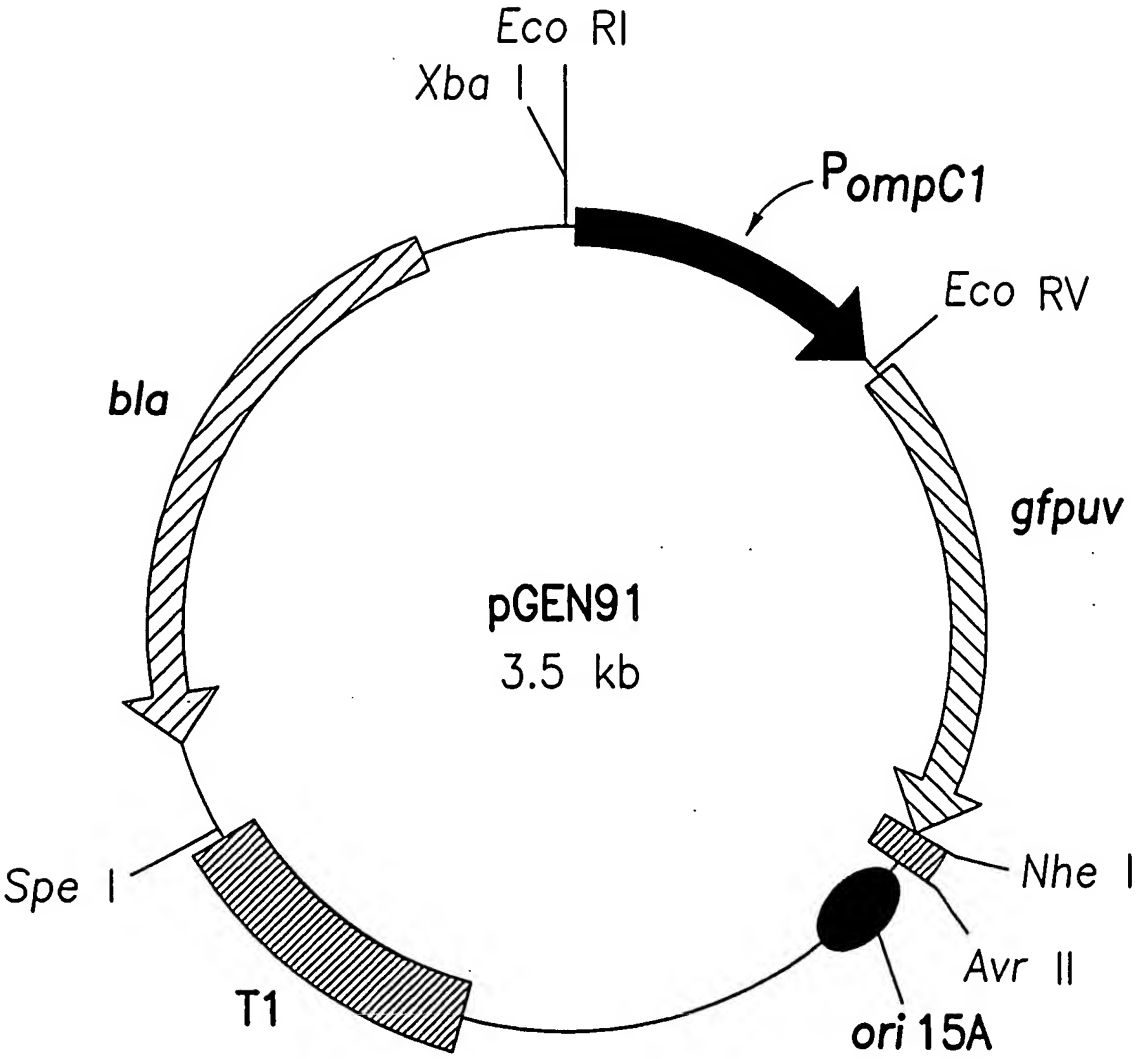

Eco RI
Xba I
PompC1
Eco RV
bla
gfpuv
pGEN91
3.5 kb
Spe I
Nhe I
Avr II
T1
ori 15A

FIG.7A

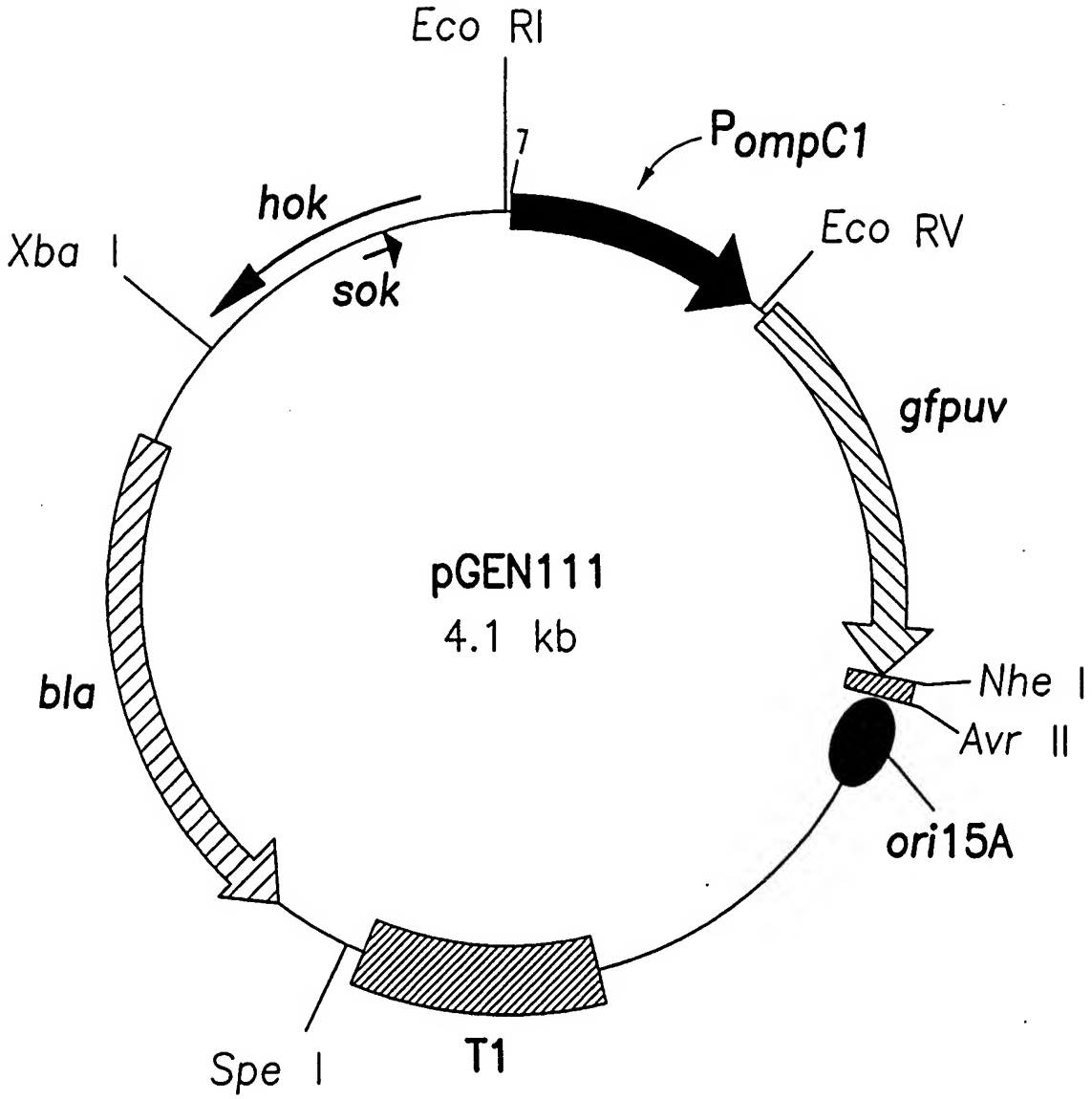

Eco RI
1
PompC1
hok
sok
Xba I
Eco RV
gfpuv
pGEN111
4.1 kb
bla
Nhe I
Avr II
ori15A
Spe I
T1

FIG.7B

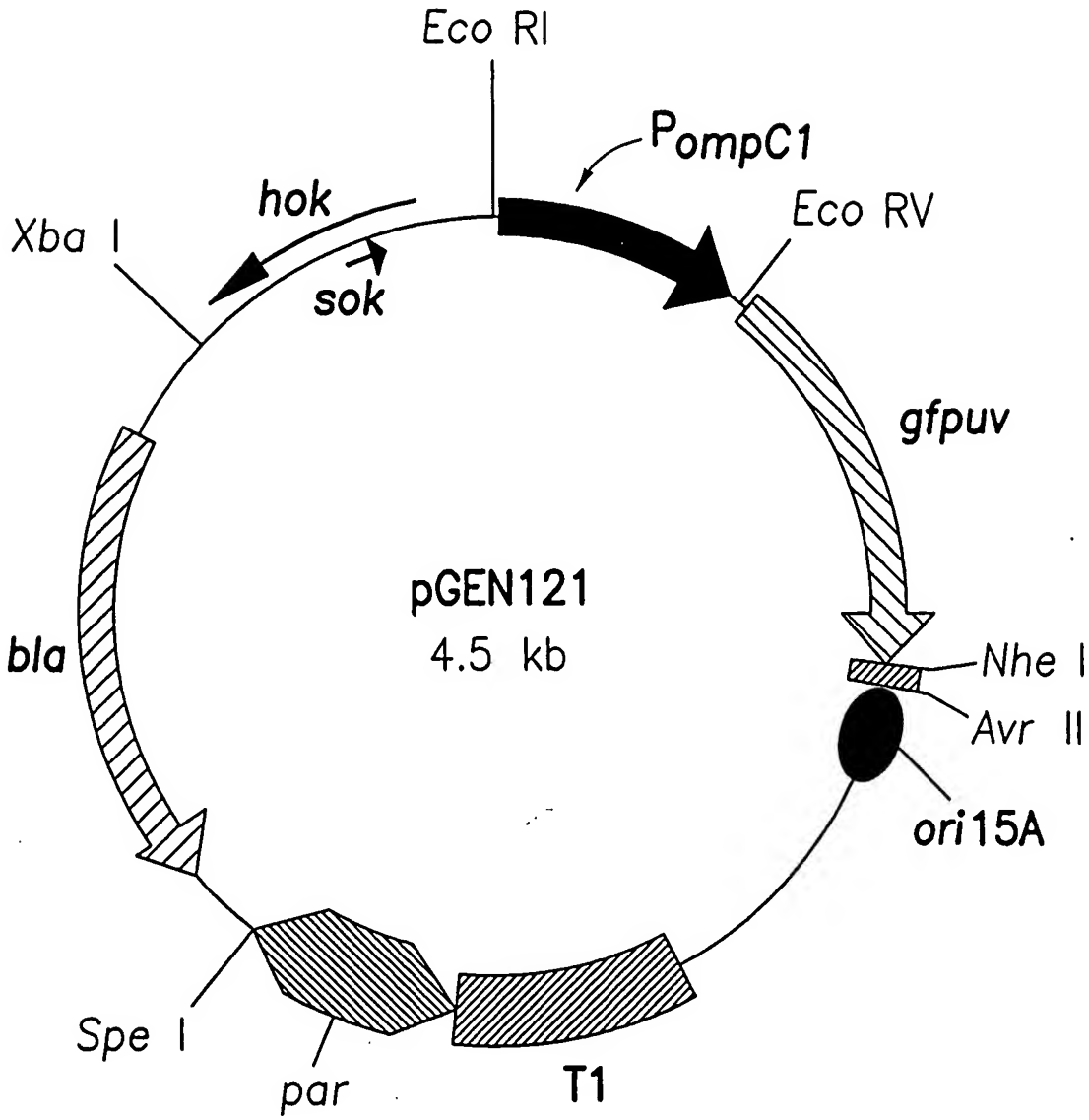

Eco RI
PompC1
hok
sok
Xba I
Eco RV
gfpuv
pGEN121
4.5 kb
bla
Nhe I
Avr II
ori15A
Spe I
par
T1

FIG.7C

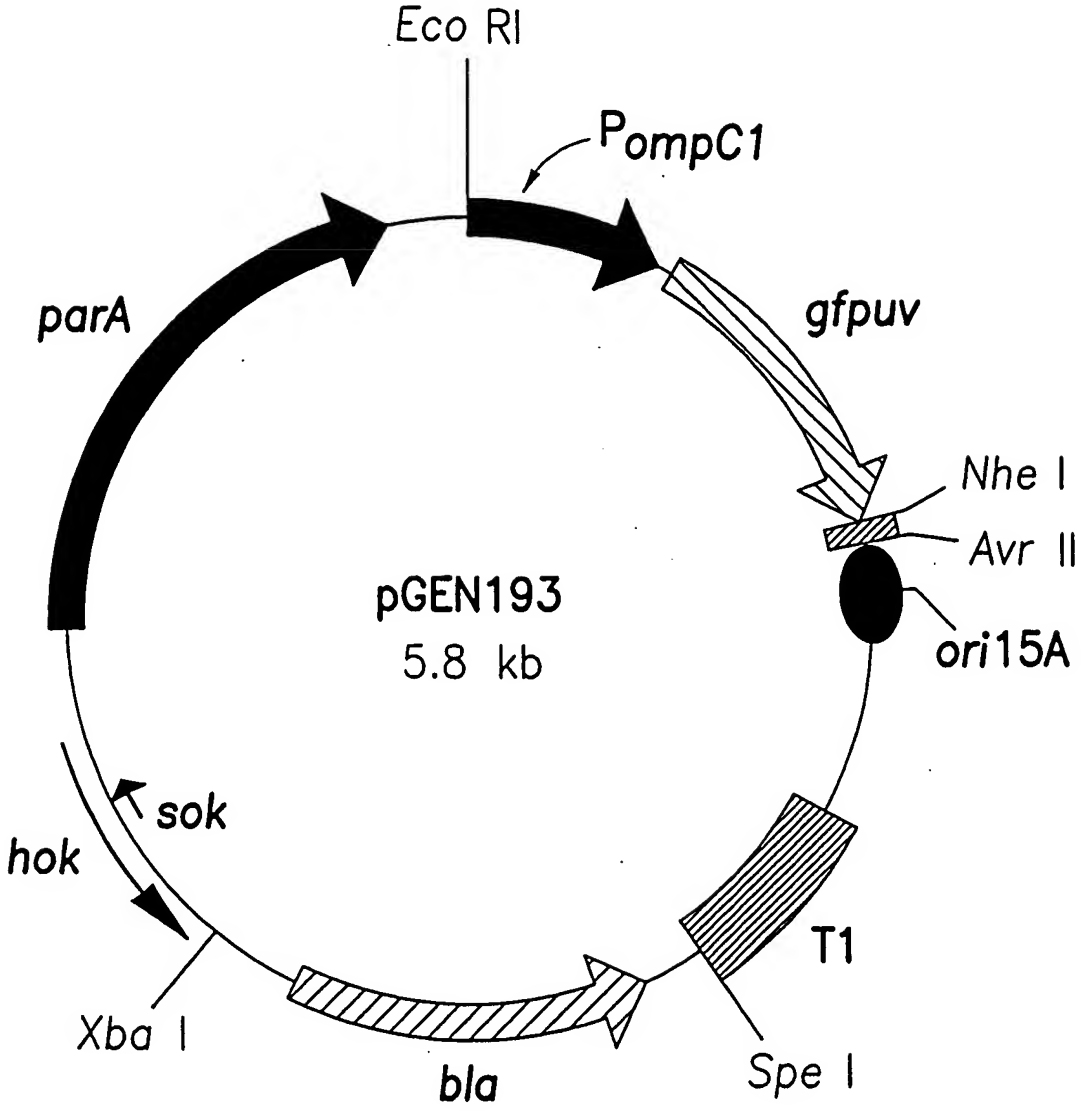

Eco RI
PompC1
gfpuv
Nhe I
Avr II
ori15A
T1
Spe I
bla
Xba I
hok
sok
parA
pGEN193
5.8 kb

FIG.7D

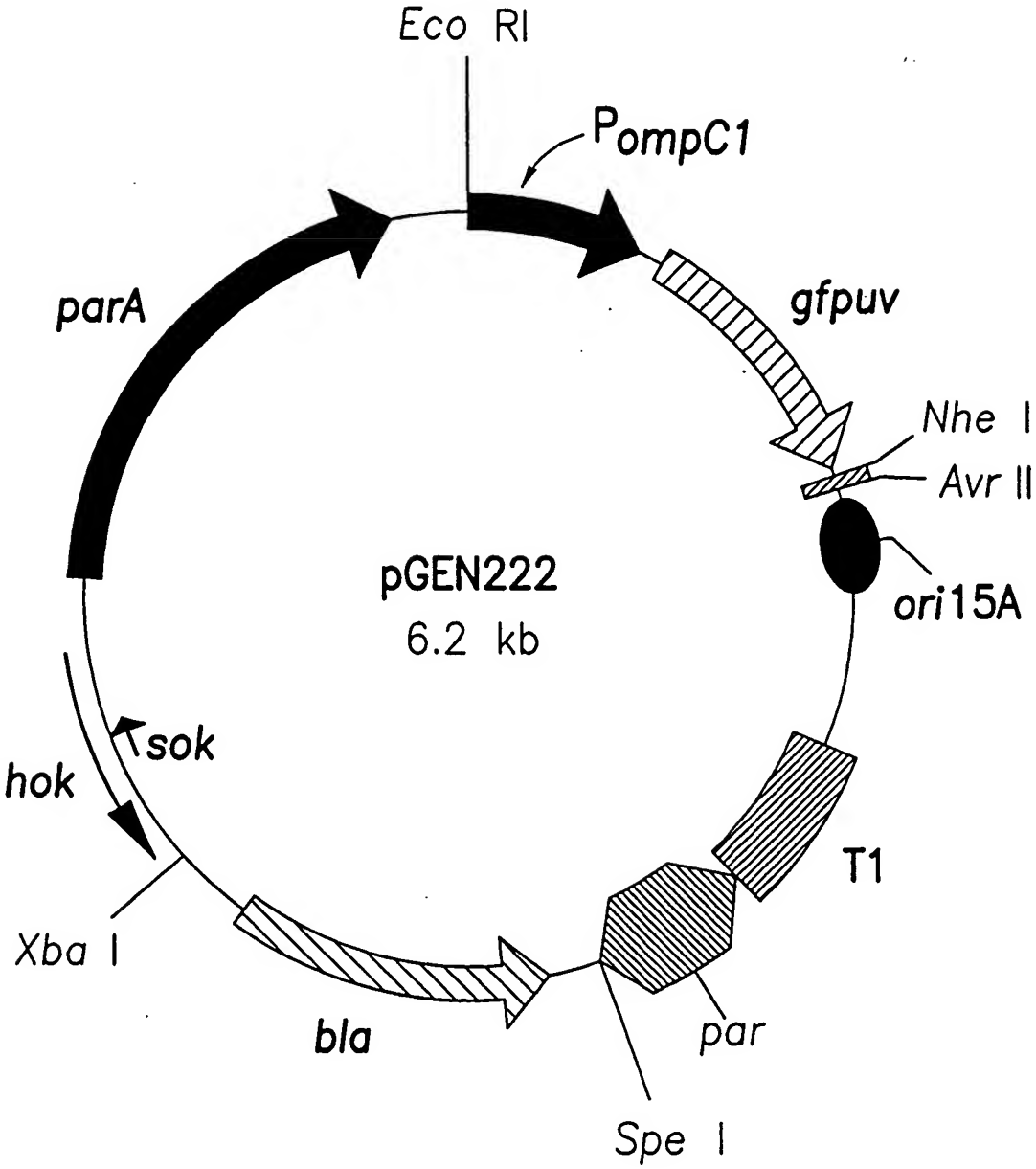
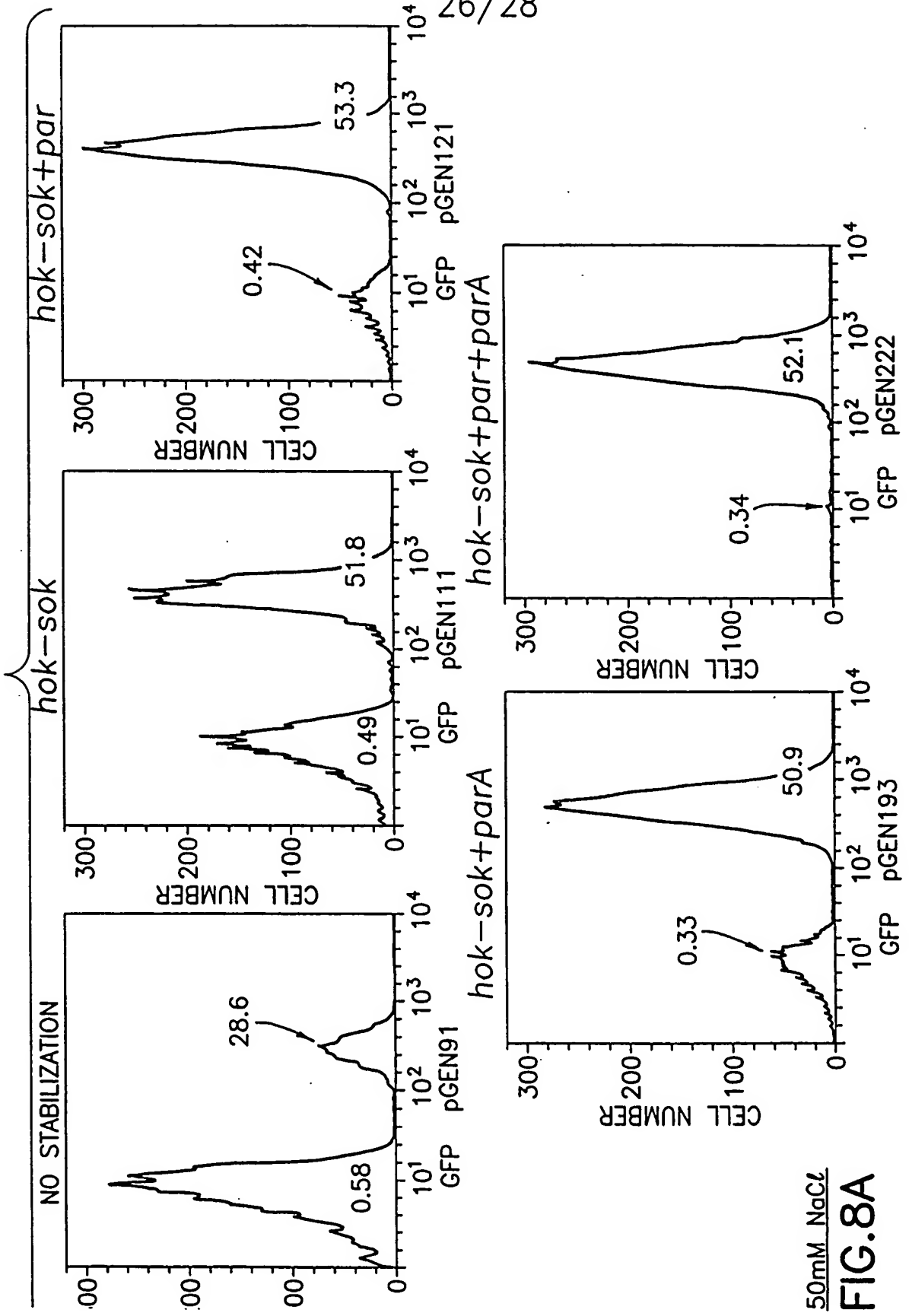

Eco RI
PompC1
gfpuv
Nhe I
Avr II
ori15A
T1
par
Spe I
bla
Xba I
hok
sok
parA
pGEN222
6.2 kb

FIG.7E



50mM NaCl

FIG.8A

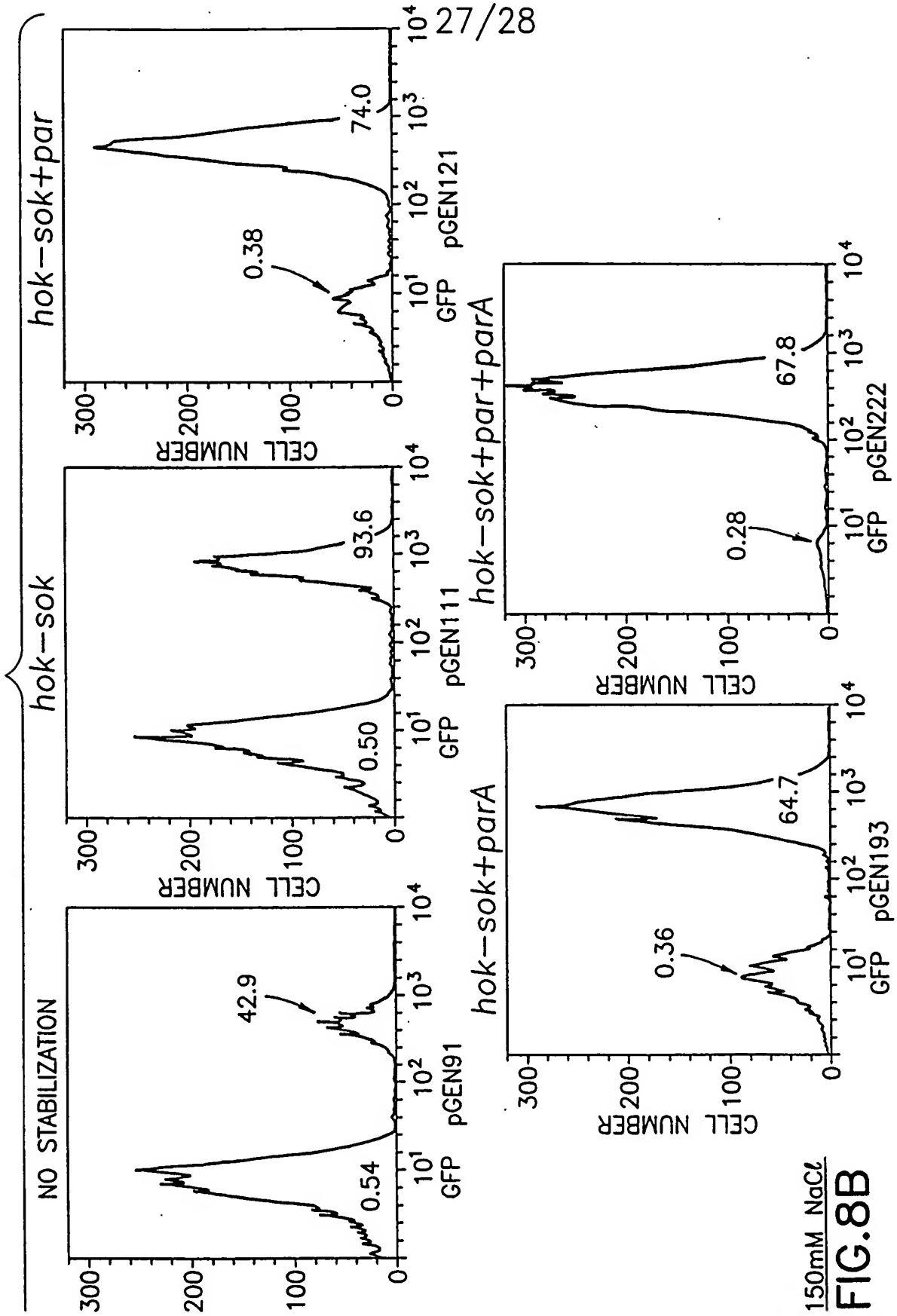
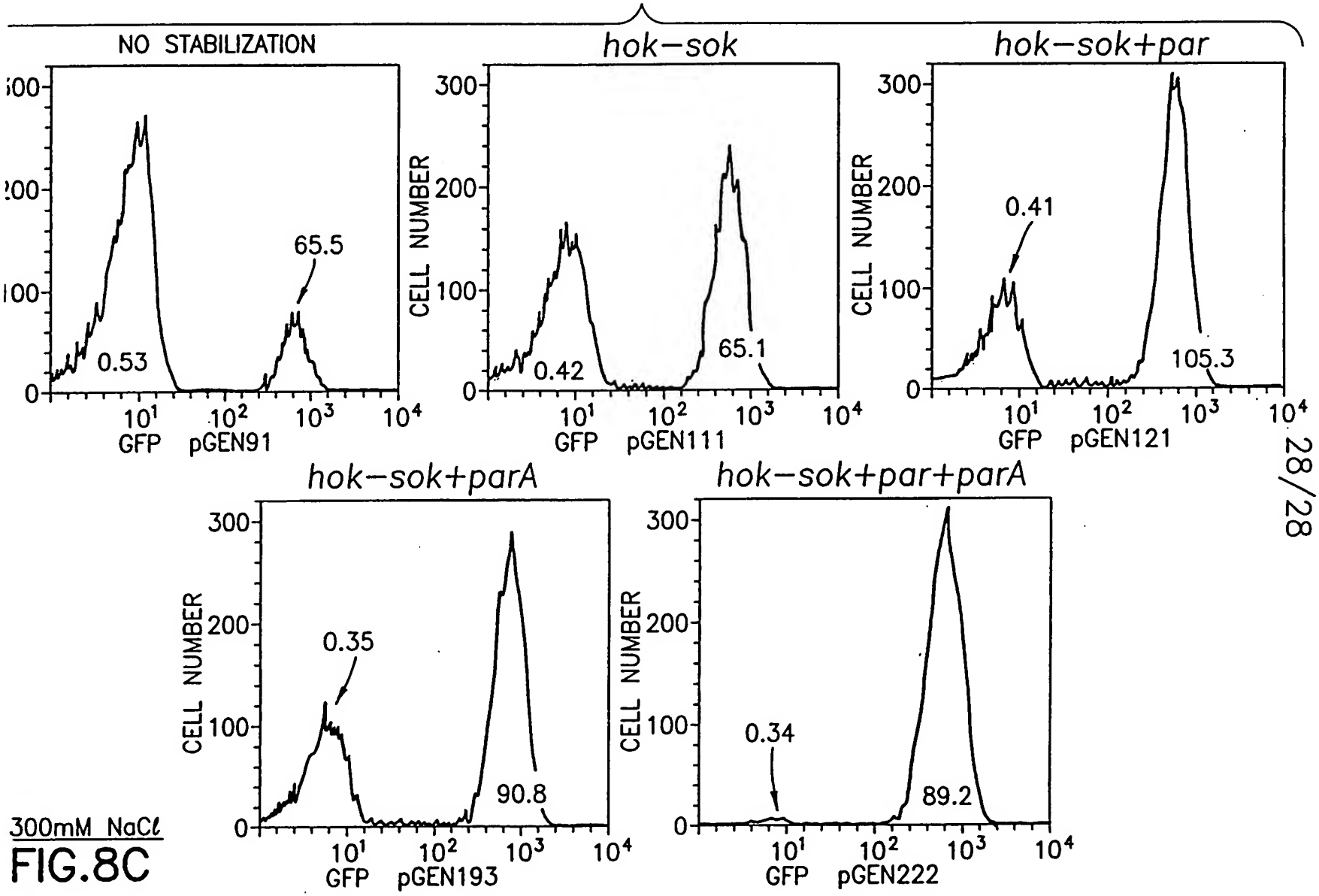

NO STABILIZATION
hok−sok
hok−sok+par
hok−sok+parA
hok−sok+par+parA
CELL NUMBER
GFP
pGEN91
pGEN111
pGEN121
pGEN193
pGEN222
42.9
0.54
93.6
0.50
74.0
0.38
64.7
0.36
67.8
0.28
150mM NaCl

FIG.8B



300mM NaCl

FIG.8C

James E. GALEN
PLASMID MAINTENANCE...
A-8703